Maternal antenatal vitamin D supplementation and offspring risk of atopic eczema in the first 4 years of life

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Abstract

Background Evidence linking prenatal maternal vitamin D supplementation and offspring risk of atopic eczema is inconsistent, with most data coming from observational studies. **Methods** Within the UK Maternal Vitamin D Osteoporosis Study (MAVIDOS) double-blind, randomised, placebo-controlled trial, we examined the relation of maternal vitamin D supplementation during pregnancy with offspring atopic eczema at ages 12, 24 and 48 months. In MAVIDOS, pregnant women were allocated to either cholecalciferol 1000 IU/day or matched placebo, taken from around 14 weeks' gestation until delivery, with the primary outcome of neonatal whole-body bone mineral content. The prevalence of atopic eczema in the offspring was ascertained at ages 12 (n=636), 24 (n=611) and 48 (n=450) months, based on the UK Working Party Criteria for the Definition of Atopic Dermatitis. **Results** Mothers and offspring characteristics were similar between the intervention and placebo groups, apart from longer breastfeeding duration in the intervention group. Adjusting for breastfeeding duration, offspring of mothers who received 1000 IU cholecalciferol daily had a lower odds ratio (OR) of atopic eczema at age 12 months: OR (95%CI) 0.55 (0.32-0.97), p=0.04. The ORs of atopic eczema in the intervention group at ages 24 and 48 months were 0.77 (0.47-1.24) and 0.71 (0.35-1.43), respectively. **Conclusion** Our data demonstrated a clinically important reduction in offspring risk of atopic eczema in infancy following maternal cholecalciferol supplementation during pregnancy. The findings support a developmental influence on infantile atopic eczema and point to gestational cholecalciferol supplementation as a preventive strategy to reduce the burden of atopic eczema during infancy.

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Background

Evidence linking prenatal maternal vitamin D supplementation and offspring risk of atopic eczema is inconsistent, with most data coming from observational studies.

Methods

Within the UK Maternal Vitamin D Osteoporosis Study (MAVIDOS) double-blind, randomised, placebo-controlled trial, we examined the relation of maternal vitamin D supplementation during pregnancy with offspring atopic eczema at ages 12, 24 and 48 months. In MAVIDOS, pregnant women were allocated to either cholecalciferol 1000 IU/day or matched placebo, taken from around 14 weeks' gestation until delivery, with the primary outcome of neonatal whole-body bone mineral content. The prevalence of atopic eczema in the offspring was ascertained at ages 12 (n=636), 24 (n=611) and 48 (n=450) months, based on the UK Working Party Criteria for the Definition of Atopic Dermatitis.

Results

Mothers and offspring characteristics were similar between the intervention and placebo groups, apart from longer breastfeeding duration in the intervention group. Adjusting for breastfeeding duration, offspring of mothers who received 1000 IU cholecalciferol daily had a lower odds ratio (OR) of atopic eczema at age 12 months: OR (95%CI) 0.55 (0.32-0.97), p=0.04. The ORs of atopic eczema in the intervention group at ages 24 and 48 months were 0.77 (0.47-1.24) and 0.71 (0.35-1.43), respectively.

Conclusion

Our data demonstrated a clinically important reduction in offspring risk of atopic eczema in infancy following maternal cholecalciferol supplementation during pregnancy. The findings support a developmental influence on infantile atopic eczema and point to gestational cholecalciferol supplementation as a preventive strategy to reduce the burden of atopic eczema during infancy.

Key words: Atopic eczema, maternal cholecalciferol, pregnancy, vitamin D

Introduction

Atopic eczema is a chronic inflammatory condition that can substantially impact affected individuals, their families and the healthcare system. The estimated prevalence of atopic eczema is 9.5% in children under age 4 years[1], with a rise observed globally over recent decades.[2] There is increasing evidence that atopic eczema partly originates *in utero* , where genetic susceptibility and environmental exposures can affect the developing immune system and alter the skin barrier. Understanding the role of early life environmental exposures, such as maternal micronutrient status, may identify potential preventative strategies.

Inadequate gestational vitamin D status is highly prevalent in many populations. Supplementation is recommended to prevent deficiency.[3] Maternal serum levels of 25(OH)D correlate with offspring levels at birth[4] and maternal vitamin D status has been extensively linked to offspring risk of atopic eczema and other atopic diseases, but with inconsistent evidence. One intervention study with high dose maternal antenatal vitamin D supplementation (2400 or 4000 IU) daily compared with placebo demonstrated a 25% reduction in the offspring's risk of 'asthma' at age 0-3 years.[5] Conversely, in an observational study, children born to mothers with a late pregnancy serum 25(OH)D >75nmol/L had a higher risk of infantile eczema at age 9 months and childhood asthma age 9 years compared to children whose mothers had a concentration of <30 nmol/l.[6]A trial in women at high risk of having offspring with asthma reported no significant difference in rates of offspring eczema at age 3 years following maternal antenatal supplementation with high (4400 IU/d) vs low (400 IU/d) dose vitamin D.[7] Maternal vitamin D supplementation during pregnancy (2000 IU/d from 27 weeks gestation) increased vitamin D activity in breast milk,[8] raising the possibility that benefits of

gestational supplementation may arise from higher infant intakes after birth in supplemented mothers who breastfeed their infant.[9]

In this study, our aim was to examine the hypothesis that maternal supplementation with 1000 IU/day cholecalciferol during pregnancy would decrease the risk of atopic eczema in the offspring in the setting of a randomised controlled trial. We also sought to determine whether any associations varied by breastfeeding status and whether genetic variants previously associated with serum 25(OH)D concentrations[10] were related to offspring atopic eczema.

Methods

Within the Maternal Vitamin D Osteoporosis Study (MAVIDOS), a multicentre, double-blind, randomised, placebo-controlled trial, women were randomised to receive cholecalciferol 1000 IU/day or matched placebo, from 14 weeks' gestation until delivery. The trial methods and primary findings have been published.[11, 12] Pregnant women were invited to participate at their early pregnancy ultrasound screening appointment. Eligible women were recruited and randomised at 14 weeks' gestation (or as soon as possible before 17 weeks' gestation if recruited later) to either cholecalciferol 1000 IU/day or matched placebo [Merck KGaA, Darmstadt, Germany)/Sharp Clinical Services (previously DHP-Bilcare), Crickhowell, UK], taken until delivery. Inclusion criteria were women aged over 18 years, having a singleton pregnancy with a gestational age <17 weeks based on last menstrual period and ultrasound measurements, and serum 25(OH)D between 25 and 100nmol/l and calcium <2.75mmol/l. Due to an ethics committee stipulation, only women with a baseline 25(OH)D between 25-100nmol/l were eligible to participate. Women were excluded if they had known metabolic bone disease, renal stones, hyperparathyroidism or hypercalciuria; were taking medication known to interfere with fetal growth or more than 400 IU/day vitamin D supplementation, or if their fetus had a major anomaly. All participants received standard antenatal care, and could continue self-administration of dietary supplements containing up to 400 IU/day vitamin D.

The MAVIDOS trial was conducted at three UK study sites 2008-2014: Southampton, Oxford and Sheffield, with a total of 965 births.[11] Follow up for this study was restricted to 704 Southampton children (352 intervention group and 352 placebo (Supplementary Figure 1)), who were assessed for eczema at ages 12 (n=636), 24 (n=611) and 48 (n=450) months. Case definition was based on the UK Working Party diagnostic criteria for the definition on atopic eczema,[13] assessed by trained research nurses who undertook a standardised questionnaire and examination. Itchy skin condition in the past 12 months was a mandatory criterion in addition to three of: onset age <2 years, history of eczema (flexural or of cheeks and extensors in under 18 months), history of dry skin the last year, and visible flexural eczema (or visible eczema of the cheeks and extensors if under 18 months). A personal history of atopy was omitted as a criterion given the young age of the infants, who were not old enough to have developed clearly defined asthma or hay fever.

The trial was approved by the Southampton and South West Hampshire Research Ethics Committee. MAVIDOS was registered prospectively (International Standard Randomised Controlled Trial Registry: ISRCTN 82927713; European Clinical Trials Database: EudraCT 2007-001716-23). Written informed consent was obtained from all parents.

Statistical analysis

Participant characteristics are described separately for mothers who received 1000 IU cholecalciferol and those who received placebo using frequency and percentage distribution for categorical variables, mean (SD) for normally distributed continuous variables and median (interquartile range) for non-normally distributed continuous variables.

We used logistic regression to examine associations between being randomised to the active group and developing eczema at ages 12, 24 and 48 months, expressing results as odds ratios (ORs) and 95% confidence intervals (CIs). Models were adjusted for duration of breastfeeding as descriptive analyses showed differences in breastfeeding duration between the groups. Sensitivity analyses were undertaken to examine whether the effect of the intervention differed in mothers breastfeeding for <1 and [?]1 month because of its reported

association with eczema and the influence of gestational supplementation on breast milk vitamin D content, and to take account of season of birth. We additionally used logistic regression to examine the associations between the offspring's risk of atopic eczema at ages 12, 24 and 48 months and maternal late pregnancy serum 25(OH)D concentration and single-nucleotide polymorphisms (SNPs) in or near key vitamin D metabolism genes, specifically rs12785878 (encoding 7-dehydrocholesterol reductase (*DHCR7)* in the epidermal vitamin D biosynthesis pathway), rs10741657 and rs6013897 (encoding 25-hydroxylase (*CYP2R1)* and 24-hydroxylase *(CYP24A1)* , respectively).[10] Analyses were performed using Stata (Stata version 15.1, Statacorp LP, TX).

Results

Cohort characteristics

Mother and infant characteristics of the 704 offspring (352 intervention group and 352 placebo) with data on atopic eczema at any of ages 12 (n=636), 24 (n=611) or 48 (n=450) months were similar in the intervention and placebo groups with the exception of longer breastfeeding duration in the intervention group (Table 1). Mother and infant characteristics apart from breastfeeding duration were also similar for the subgroups followed up at each of the three postnatal ages (Supplementary Table 1), and the 704 mothers and offspring included had characteristics similar to those of the overall group recruited to MAVIDOS (Supplementary Table 2). Baseline maternal serum 25(OH)D levels at recruitment in early pregnancy were similar in the intervention and placebo groups. In late pregnancy, maternal serum 25(OH)D levels were higher in the intervention group compared with the placebo group (Table 1).

Association between maternal cholecalciferol supplementation and offspring atopic eczema

The prevalences of atopic eczema in the intervention group at ages 12, 24 and 48 months were 7.2%, 11.4% and 6.7% respectively, compared with 12.0%, 14.5% and 8.8% in the placebo group. Table 2 shows the OR of atopic eczema at ages 12, 24 and 48 months in offspring whose mothers received 1000IU cholecalciferol vs placebo. In an unadjusted analysis, offspring of mothers who received 1000IU cholecalciferol had a lower risk OR of atopic eczema at age 12 months, 0.57 (95%CI 0.33, 0.98), p = 0.04 compared to offspring of mothers who received placebo; this changed little after adjusting for breastfeeding duration as a covariate. The ORs of atopic eczema in the intervention group compared to the control group at ages 24 and 48 months were 0.75 (0.47, 1.21) p = 0.25, and 0.75 (0.37, 1.50) p = 0.41, respectively, and changed little in models adjusted for breastfeeding (Table 2) or season at birth. Adjusted estimates are presented graphically in Figure 1. Sensitivity analysis stratified by breastfeeding duration demonstrated a reduced risk of atopic eczema at age 12 months in the intervention group in infants who were breastfed for more than one month (OR 0.48 (0.24,0.94), p = 0.03), but not in those who were breastfed for less than one month (OR 0.80 (0.29,2.17), p = 0.66) (Table 3); however, interaction terms between the intervention and breastfeeding duration were not statistically significant at any of the three follow-up ages (p = 0.41, 0.35, 0.13 at ages 12, 24 and 48 months, respectively).

Sensitivity analyses showed no association between maternal late pregnancy serum 25(OH)D and offspring atopic eczema at any age (Supplementary table 3). Examining the SNPs rs12785878 (*DHCR7*), rs10741657 (*CYP2R1*) and rs6013897 (*CYP24A1*) located in genes involved in vitamin D metabolism[10, 14], demonstrated no associations with offspring atopic eczema, although there indications of higher ORs for atopic eczema with rs10741657 (*CYP2R1*) at ages 12 and 24 months, OR 1.44 (0.95, 2.18), p = 0.08, and 1.42 (0.98, 2.04), p = 0.06, respectively (Table 4).

Discussion

Maternal supplementation with 1000 IU cholecalciferol daily from 14 weeks' gestation until delivery resulted in a reduced odds of infant atopic eczema at age 12 months. The effects of supplementation, however, were non-significant at ages 24 and 48 months. Interaction terms between supplementation during pregnancy and breastfeeding duration were not statistically significant, but sensitivity analysis showed that the protective effect of maternal cholecalciferol supplementation on infantile eczema was only significant in offspring breastfed for more than one month.

Current evidence relating to maternal vitamin D status and its effect on offspring atopic eczema is inconsistent, and evidence from supplementation trials is sparse. A U-shaped association between maternal vitamin D supply and status and offspring atopic eczema is biologically plausible, whereby both low and high intakes and 25(OH)D insufficiency and high 25(OH)D concentrations might be associated with increased risk of atopic eczema (see below for discussion of potential mechanisms).[6] We would note that mothers with serum 25(OH)D levels above 100 mmol/L at baseline were excluded from MAVIDOS, but found no evidence for an increased risk of atopic eczema with 1000 IU/d cholecalciferol supplementation. A reduced risk of wheeze and eczema has been reported in children of mothers consumed 174 IU/d or more of dietary vitamin D during pregnancy[15], and infants with cord blood 25(OH)D levels [?] 75 nmol/L were found to have a lower risk of eczema in infancy when compared to those with cord blood levels of $<$ 50 nmol/L.[16] There are also reports of no association between maternal or cord serum 25(OH)D concentrations and atopic eczema.[17] Others studies have reported no clear associations between maternal vitamin D status in late pregnancy and asthma, wheeze or skin sensitisation at age 1, 3 or 6 years.[18]

In the VDAART randomised controlled trial in women at high risk of having children with asthma, the prevalence of offspring asthma at age 6 years was similar in those whose mothers received antenatal supplementation with 4400 vs 400 IU/d vitamin D, as were the secondary outcomes of eczema and total IgE levels; a between-group reduction in asthma and recurrent wheeze was, however, suggested at early time points through the age of 3 years.[7, 19] Our study examined the effect of gestational supplementation on offspring eczema at ages from 12 to 48 months, also finding an effect at early age, 12 months. We found no effect on eczema in the past year at ages 24 and 48 months suggesting that other, post-natal influences might become important at older ages in affecting the risk of atopic eczema beyond infancy. Conceivably, supplementation during the postnatal period may be needed for a sustained effect. There is evidence supportive of postnatal vitamin D supplementation, with a meta-analysis of 11 intervention studies in children with atopic eczema reporting a reduction in eczema severity.[20]

Vitamin D has immunomodulatory effects on innate and adaptive responses.[21] The vast majority of cells of the adaptive immune system express the vitamin D receptor and CYP27B1, enabling the production of the active metabolite 1,25(OH)2 D, thought to act predominantly in an auto-and paracrine fashion. Evidence from*in vitro* and *in vivo* studies[22] has demonstrated that vitamin D supplementation inhibits expression of Th2 response cytokines, the predominant immune response seen acutely in atopic eczema and other allergic disease. [23]Vitamin D deficiency *in utero* and early life has been linked with increased Th2 lymphocytes and reduced T regulatory cells and interleukin IL-10, leading to macrophage and dendritic cells producing pro-inflammatory cytokines.[24] However, contrary to this, there is evidence that 1,25(OH)2 D promotes Th2 responses, with inhibition of IFN-γ and promotion of IL-4, IL-5, and IL-10 production.[25] Skin barrier function is important in the pathogenesis of atopic eczema; vitamin D and its metabolites can impact this through involvement in the synthesis of proteins such as filaggrin, and through stratum corneum formation, keratinocyte formation and differentiation and production and the regulation of skin antimicrobial peptides.[26]

Our data suggest that the effect of vitamin D supplementation on offspring eczema risk may be seen soon after pregnancy, but weaken as children grow older, where other risk factors can be influential. We speculate that during infancy there may be a role of breast milk vitamin D content. Evidence from MAVIDOS has demonstrated an increase in maternal serum levels with 1000 IU/d cholecalciferol supplementation,[12] but in line with the Southampton Women's Survey observational study,[18] our data showed no association between maternal serum 25(OH)D in late pregnancy and offspring atopic eczema. Previous studies have shown that gestational vitamin D supplementation increases breast milk vitamin D content,[8, 9] and in MAVIDOS the vitamin D content of breast milk is likely to have been higher in the supplemented group, influenced by mobilisation from maternal fat and muscle tissue. This may explain our finding of a protective effect only in children that were breastfed for more than 1 month. Heterogeneity in the aetiology and pathogenesis of atopic eczema in early childhood is increasingly recognised,[27] and an alternative possibility is that vitamin D supplementation may only have an effect on particular atopic eczema phenotypes.

In MAVIDOS, examination of genetic variants in genes related to the vitamin D pathway has shown that rs12785878 (DHCR7) was associated with baseline 25(OH)D, likely influencing cutaneous synthesis; achieved 25(OH)D status following supplementation was associated with rs10741657 (CYP2R1, which determines the efficiency of vitamin D to 25(OH)D conversion), whereas rs12785878 and rs6013897 (CYP24A1) were not.[10] There were weak trends for higher ORs of atopic eczema with rs10741657 (CYP2R1) at ages 12 and 24 months but no associations for other SNPs examined. In a case-control study of Chinese children, rs4674343 on CYP27A1 (27-hydroxylase, an enzyme converting the pre-vitamin D3 metabolite lumisterol into further downstream metabolites with biological activity in skin cells[28]) was reported to be protective against atopic eczema and CYP2R1 and VDR haplotypes also influenced atopic eczema risk and eosinophil count.[29]

A strength of this study is analysis of data from a placebo-controlled, double-blind, randomised trial. Atopic eczema was not the primary outcome in MAVIDOS, but the data collected enabled ascertainment of offspring atopic eczema using the UKWPDC well-recognised criteria for the diagnosis of atopic eczema. Furthermore, these criteria were determined by trained research nurses who examined the offspring. While some participants were taking vitamin D in addition to the intervention/placebo provided, supplement use at interview did not differ between the intervention and placebo groups. A limitation of this study is that cord blood and offspring 25(OH)D levels were not measured, precluding examination of these in relation to atopic eczema. Possible effects of UVB and any interaction with supplementation could not be investigated because no UVB exposure data were available. Our study population did not include many women who were not white and we could not include participants with baseline 25(OH)D concentrations of less than 25 nmol/L as a result of stipulations made during the ethics approval process; these reduce the generalisability of our findings.

In conclusion, in a randomised controlled trial maternal supplementation with 1000 IU/d cholecalciferol from 14 weeks' gestation to delivery led to a reduced incidence of atopic eczema in the first year of life. Many international and national guidelines recommend 400-600 IU/d (10-15 micrograms) cholecalciferol throughout pregnancy, with the strongest evidence for the prevention of neonatal hypocalcaemia and emerging evidence for effects on other health outcomes affecting skeletal, respiratory and immune systems.[30] The current findings inform understanding of the early life influences on infantile eczema and support recommendations for routine vitamin D supplementation during pregnancy.

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Competing interests

KMG has received reimbursement for speaking at conferences sponsored by companies selling nutritional products. KMG is part of an academic consortium that has received research funding from Abbott Nutrition, Nestec and Danone. CC reports personal fees from ABBH, Amgen, Eli Lilly, GSK, Medtronic, Merck,

Novartis, Pfizer, Roche, Servier and Takeda, outside the submitted work. NCH reports personal fees, consultancy, lecture fees and honoraria from Alliance for Better Bone Health, AMGEN, MSD, Eli Lilly, Servier, Shire, Radius Health, UCB, Consilient Healthcare and Internis Pharma, outside the submitted work. The other authors have no competing interests.

References

1. Simpson CR, Anderson WJ, Helms PJ, Taylor MW, Watson L, Prescott GJ, et al. Coincidence of immune-mediated diseases driven by Th1 and Th2 subsets suggests a common aetiology. A population-based study using computerized general practice data. Clin Exp Allergy. 2002;32(1):37-42.

2. Asher MI, Montefort S, Bjorksten B, Lai CK, Strachan DP, Weiland SK, et al. Worldwide time trends in the prevalence of symptoms of asthma, allergic rhinoconjunctivitis, and eczema in childhood: ISAAC Phases One and Three repeat multicountry cross-sectional surveys. Lancet. 2006;368(9537):733-43.

3. Moon RJ, Harvey NC, Cooper C, D'Angelo S, Crozier SR, Inskip HM, et al. Determinants of the Maternal 25-Hydroxyvitamin D Response to Vitamin D Supplementation During Pregnancy. The Journal of clinical endocrinology and metabolism. 2016;101(12):5012-20.

4. Maghbooli Z, Hossein-Nezhad A, Shafaei AR, Karimi F, Madani FS, Larijani B. Vitamin D status in mothers and their newborns in Iran. BMC Pregnancy Childbirth. 2007;7:1.

5. Maciag MC, Phipatanakul W. Preventing the development of asthma: stopping the allergic march. Curr Opin Allergy Clin Immunol. 2019;19(2):161-8.

6. Gale CR, Robinson SM, Harvey NC, Javaid MK, Jiang B, Martyn CN, et al. Maternal vitamin D status during pregnancy and child outcomes. Eur J Clin Nutr. 2008;62(1):68-77.

7. Litonjua AA, Carey VJ, Laranjo N, Harshfield BJ, McElrath TF, O'Connor GT, et al. Effect of Prenatal Supplementation With Vitamin D on Asthma or Recurrent Wheezing in Offspring by Age 3 Years: The VDAART Randomized Clinical Trial. JAMA. 2016;315(4):362-70.

8. Wall CR, Stewart AW, Camargo CA, Jr., Scragg R, Mitchell EA, Ekeroma A, et al. Vitamin D activity of breast milk in women randomly assigned to vitamin D3 supplementation during pregnancy. Am J Clin Nutr. 2016;103(2):382-8.

9. Jan Mohamed HJ, Rowan A, Fong B, Loy SL. Maternal serum and breast milk vitamin D levels: findings from the Universiti Sains Malaysia Pregnancy Cohort Study. PLoS One. 2014;9(7):e100705.

10. Moon RJ, Harvey NC, Cooper C, D'Angelo S, Curtis EM, Crozier SR, et al. Response to Antenatal Cholecalciferol Supplementation Is Associated With Common Vitamin D-Related Genetic Variants. J Clin Endocrinol Metab. 2017;102(8):2941-9.

11. Harvey NC, Javaid K, Bishop N, Kennedy S, Papageorghiou AT, Fraser R, et al. MAVIDOS Maternal Vitamin D Osteoporosis Study: study protocol for a randomized controlled trial. The MAVIDOS Study Group. Trials. 2012;13:13.

12. Cooper C, Harvey NC, Bishop NJ, Kennedy S, Papageorghiou AT, Schoenmakers I, et al. Maternal gestational vitamin D supplementation and offspring bone health (MAVIDOS): a multicentre, double-blind, randomised placebo-controlled trial. Lancet Diabetes Endocrinol. 2016;4(5):393-402.

13. Williams HC, Burney PG, Hay RJ, Archer CB, Shipley MJ, Hunter JJ, et al. The U.K. Working Party's Diagnostic Criteria for Atopic Dermatitis. I. Derivation of a minimum set of discriminators for atopic dermatitis. Br J Dermatol. 1994;131(3):383-96.

14. Curtis EM, Moon RJ, Harvey NC, Cooper C. Maternal vitamin D supplementation during pregnancy. Br Med Bull. 2018;126(1):57-77.

15. Miyake Y, Sasaki S, Tanaka K, Hirota Y. Dairy food, calcium and vitamin D intake in pregnancy, and wheeze and eczema in infants. Eur Respir J. 2010;35(6):1228-34.

16. Jones AP, D'Vaz N, Meldrum S, Palmer DJ, Zhang G, Prescott SL. 25-hydroxyvitamin D3 status is associated with developing adaptive and innate immune responses in the first 6 months of life. Clin Exp Allergy. 2015;45(1):220-31.

17. O'Donovan SM, J OBH, Murray DM, Kenny LC, Khashan AS, Chaoimh CN, et al. Neonatal adiposity increases the risk of atopic dermatitis during the first year of life. J Allergy Clin Immunol. 2016;137(1):108-17.

18. Pike KC, Inskip HM, Robinson S, Lucas JS, Cooper C, Harvey NC, et al. Maternal late-pregnancy serum 25-hydroxyvitamin D in relation to childhood wheeze and atopic outcomes. Thorax. 2012;67(11):950-6.

19. Litonjua AA, Carey VJ, Laranjo N, Stubbs BJ, Mirzakhani H, O'Connor GT, et al. Six-Year Follow-up of a Trial of Antenatal Vitamin D for Asthma Reduction. N Engl J Med. 2020;382(6):525-33.

20. Hattangdi-Haridas SR, Lanham-New SA, Wong WHS, Ho MHK, Darling AL. Vitamin D Deficiency and Effects of Vitamin D Supplementation on Disease Severity in Patients with Atopic Dermatitis: A Systematic Review and Meta-Analysis in Adults and Children. Nutrients. 2019;11(8).

21. Szekely JI, Pataki A. Effects of vitamin D on immune disorders with special regard to asthma, COPD and autoimmune diseases: a short review. Expert Rev Respir Med. 2012;6(6):683-704.

22. Vasiliou JE, Lui S, Walker SA, Chohan V, Xystrakis E, Bush A, et al. Vitamin D deficiency induces Th2 skewing and eosinophilia in neonatal allergic airways disease. Allergy. 2014;69(10):1380-9.

23. Baker KE, Bonvini SJ, Donovan C, Foong RE, Han B, Jha A, et al. Novel drug targets for asthma and COPD: lessons learned from in vitro and in vivo models. Pulm Pharmacol Ther. 2014;29(2):181-98.

24. Cyprian F, Lefkou E, Varoudi K, Girardi G. Immunomodulatory Effects of Vitamin D in Pregnancy and Beyond. Front Immunol. 2019;10:2739.

25. Boonstra A, Barrat FJ, Crain C, Heath VL, Savelkoul HF, O'Garra A. 1alpha,25-Dihydroxyvitamin d3 has a direct effect on naive CD4(+) T cells to enhance the development of Th2 cells. J Immunol. 2001;167(9):4974-80.

26. Vestita M, Filoni A, Congedo M, Foti C, Bonamonte D. Vitamin D and atopic dermatitis in childhood. J Immunol Res. 2015;2015:257879.

27. Loo EX, Shek LP, Goh A, Teoh OH, Chan YH, Soh SE, et al. Atopic Dermatitis in Early Life: Evidence for at Least Three Phenotypes? Results from the GUSTO Study. Int Arch Allergy Immunol. 2015;166(4):273-9.

28. Tuckey RC, Li W, Ma D, Cheng CYS, Wang KM, Kim T-K, et al. CYP27A1 acts on the pre-vitamin D3 photoproduct, lumisterol, producing biologically active hydroxy-metabolites. The Journal of Steroid Biochemistry and Molecular Biology. 2018;181:1-10.

29. Wang SS, Hon KL, Kong AP, Pong HN, Wong GW, Leung TF. Vitamin D deficiency is associated with diagnosis and severity of childhood atopic dermatitis. Pediatr Allergy Immunol. 2014;25(1):30-5.

30. Harvey NC, Holroyd C, Ntani G, Javaid K, Cooper P, Moon R, et al. Vitamin D supplementation in pregnancy: a systematic review. Health Technol Assess. 2014;18(45):1-190.

Table 1: Characteristics of mothers and offspring with eczema data at age 12, 24 or 48 months. (n=704)

	Placebo	**Cholecalciferol (1000 IU/da**
N	352	352
Maternal characteristics		

	Placebo	**Cholecalciferol (1000 IU/da**
Age (years), mean (SD)	31.1 (5.0)	31.0 (4.9)
Ethnicity, % Caucasian	95.8%	95.2%
Parity, % nulliparous	43.0%	44.1%
Smoking in early pregnancy, %	6.9%	5.3%
Educational attainment [?] A level, %	77.0%	78.9%
BMI (kg/m^2), median (IQR)	25.4 (22.8,29.4)	24.6 (22.2,28.2)
Sum of all skinfold thicknesses (mm), mean (SD)	81.3 (27.2)	77.8 (28.4)
Early pregnancy 25(OH)D (nmol/l), mean (SD)	44.7 (16.2)	46.0 (16.4)
Late pregnancy 25(OH)D (nmol/l), mean (SD)	42.4 (20.8)	67.4 (19.9)
Change in 25(OH)D from early to late pregnancy (nmol/l), mean (SD)	-2.0 (20.7)	21.4 (21.9)
Offspring characteristics		
Male, (%)	50.1%	55.7%
Birth weight (g), mean (SD)	3543.3 (494.9)	3508.9 (536.2)
Age last breastfed (months), median (IQR)	4.0 (0,9.0)	5.0 (1.0,10.0)

Table 2. Offspring atopic eczema in the intervention group compared to the control group

	Unadjusted	Unadjusted	Unadj
Outcome: Atopic eczema	N	OR (95%CI)	p
12 months	636	0.57 (0.33,0.98)	0.04
24 months	611	0.75 (0.47,1.21)	0.25
48 months	450	0.75 (0.37,1.50)	0.41
adjusted for breastfeeding duration	* adjusted for breastfeeding duration	* adjusted for breastfeeding duration	

Table 3. Offspring atopic eczema in the intervention group, stratified by breastfeeding duration

	Breastfed up to 1 month	Breastfed up to 1 month	Breastfed up to 1 month	Breastfed mor
Outcome: Atopic eczema	N	OR (95%CI)	p	N
12 months	248	0.80 (0.29,2.17)	0.66	374
24 months	227	1.03 (0.47,2.27)	0.95	370
48 months	164	1.69 (0.44,6.52)	0.45	275

Table 4. SNP associations with offspring atopic eczema at ages 12, 24 and 48 months

SNP	**Reference allele**	**Univariate**	**Univariate**	**Univariate**	**Adjusted for breastfe**
		n	**OR* (95%CI)**	**p**	**n**
Atopic eczema at 12 months					
rs12785878 (*DHCR7*)	G	621	0.86 (0.56,1.32)	0.48	607
rs10741657 (*CYP2R1*)	A	613	1.42 (0.95,2.12)	0.09	599
rs6013897 (*CYP24A1*)	A	615	0.78 (0.50,1.22)	0.27	601
Atopic eczema at 24 months					
rs12785878 (*DHCR7*)	G	594	1.00 (0.67,1.50)	0.99	580
rs10741657 (*CYP2R1*)	A	587	1.37 (0.96,1.97)	0.09	573
rs6013897 (*CYP24A1*)	A	590	0.95 (0.62,1.46)	0.83	576
Atopic eczema at 48 months					

SNP	Reference allele	Univariate	Univariate	Univariate	Adjusted for breastfe
rs12785878 (*DHCR7*)	G	438	1.29 (0.68,2.45)	0.43	428
rs10741657 (*CYP2R1*)	A	430	1.10 (0.66,1.83)	0.71	420
rs6013897 (*CYP24A1*)	A	434	0.85 (0.47,1.56)	0.61	424

*OR are per risk allele

Figure legends

Figure 1. Offspring odds ratios of atopic eczema at ages 12, 24 and 48 months in the intervention group whose mothers received 1000IU cholecalciferol daily during pregnancy vs placebo group.

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